19. G. D. Rochester and C. C. Butler, *Nature* **160**, 855 (1947).
20. A. J. Seriff, R. B. Leighton, C. Hsiao, E. D. Cowan, C. D. Anderson, *Phys. Rev.* **78**, 290 (1950).
21. R. Armenteros, K. H. Barker, C. C. Butler, A. Cachon, C. M. York, *Phil. Mag.* **43**, 597 (1952).
22. L. Leprince-Ringuet and M. L' Héritier, *Compt. Rend.* **219**, 618 (1944).
23. H. A. Bethe, *Phys. Rev.* **70**, 821 (1946).
24. R. M. Brown, U. Camerini, P. H. Fowler, H. Muirhead, C. F. Powell, D. M. Ritson, *Nature* **163**, 47 (1949).
25. C. O'Ceallaigh, *Phil. Mag.* **42**, 1032 (1951).
26. M. G. K. Menon and C. O'Ceallaigh, *Proc. Roy. Soc.* (*London*) **221A**, 292 (1954).
27. R. W. Thompson, A. V. Buskirk, L. R. Etter, C. J. Karzmark, R. H. Rediker, *Phys. Rev.* **90**, 329 (1953).
28. A. Bonetti, R. Levi Setti, M. Panetti, G. Tomasini, *Nuovo Cimento* **10**, 345 (1953).
29. R. Marshak and H. Bethe, *Phys. Rev.* **72**, 506 (1947).
30. M. Gell-Mann, *ibid.* **92**, 833 (1953).
31. K. Nishijima, *Progr. Theoret. Phys.* (*Kyoto*) **12**, 107 (1954).
32. L. W. Alvarez and S. Goldhaber, *Nuovo Cimento* **2**, 344 (1955).
33. T. O. Lee, Les Prix Nobel en 1957.
34. C. N. Yang, Les Prix Nobel en 1957.
35. L. W. Alvarez, F. S. Crawford, Jr., M. L. Good, M. L. Stevenson, *Phys. Rev.* **101**, 303 (1956).
36. V. Fitch and R. Motley, *ibid.*, p. 496.
37. S. von Friesen, *Arkiv. Fysik* **8**, 309 (1954); *ibid.* **10**, 460 (1956).
38. R. W. Birge, D. H. Perkins, J. R. Peterson, D. H. Stork, M. N. Whitehead, *Nuovo Cimento* **4**, 834 (1956).
39. D. Glaser, Les Prix Nobel en 1960.
40. R. H. Hildebrand and D. E. Nagle, *Phys. Rev.* **92**, 517 (1953).
41. J. G. Wood, *ibid.* **94**, 731 (1954).
42. D. P. Parmentier and A. J. Schwemin, *Rev. Sci. Instr.* **26**, 958 (1955).
43. D. C. Gates, R. W. Kenney, W. P. Swanson, *Phys. Rev.* **125**, 1310 (1962).
44. L. W. Alvarez, F. S. Crawford, Jr., M. L. Stevenson, *ibid.* **112**, 1267 (1958).
45. L. W. Alvarez, P. Davey, R. Hulsizer, J. Snyder, A. J. Schwemin, R. Zane, Lawrence Radiation Laboratory Report UCRL-10109, 1962 (unpublished); P. G. Davey, R. I. Hulsizer, W. E. Humphrey, J. H. Munson, R. R. Ross, A. J. Schwemin, *Rev. Sci. Instr.* **35**, 1134 (1964).
46. L. W. Alvarez, in *Proceedings of the 1966 International Conference on Instrumentation for High Energy Physics, Stanford, California*, p. 271.
47. W. K. H. Panofsky, L. Aamodt, H. F. York, *Phys. Rev.* **78**, 825 (1950).
48. L. W. Alvarez, H. Bradner, P. Falk-Vairant, J. D. Gow, A. H. Rosenfeld, F. T. Solmitz, R. D. Tripp, *Nuovo Cimento* **5**, 1026 (1957).
49. L. W. Alvarez, H. Bradner, F. S. Crawford, Jr., J. A. Crawford, P. Falk-Vairant, M. L. Good, J. D. Cow, A. H. Rosenfeld, F. T. Solmitz, M. L. Stevenson, H. K. Ticho, R. D. Tripp, *Phys. Rev.* **105**, 1127 (1957).
50. F. C. Frank, *Nature* **160**, 525 (1947).
51. Ya. B. Zel'dovitch, *Dokl. Akad. Nauk SSSR* **95**, 493 (1954).
52. C. S. Wu, E. Ambler, R. W. Hayward, D. D. Hoppes, R. P. Hudson, *Phys. Rev.* **105**, 1413 (1957).
53. T. D. Lee and C. N. Yang, *ibid.* **104**, 254, 822 (1956).
54. F. S. Crawford, Jr., M. Cresti, M. L. Good, K. Gottstein, E. M. Lyman, F. T. Solmitz, M. L. Stevenson, H. K. Ticho, *ibid.* **108**, 1102 (1957).
55. P. Eberhard, M. L. Good, H. Ticho, Lawrence Radiation Laboratory Report UCRL-8878, Aug. 1959 (unpublished); J. J. Murray, *ibid.* UCRL-3492, May 1957 (unpublished); ———, *ibid.* UCRL-9506, Sept. 1960 (unpublished).
56. R. H. Dalitz, *Phil. Mag.* **44**, 1068 (1953).
57. W. F. Cartwright, C. Richman, M. N. Whitehead, H. A. Wilcox, *Phys. Rev.* **78**, 823 (1950); D. L. Clark, A. Roberts, R. Wilson, *ibid.* **83**, 649 (1951); R. Durbin, H. Loar, J. Steinberger, *ibid.*, p. 646.
58. M. E. L. Oliphant and E. Rutherford, *Proc. Roy. Soc.* (*London*) **141A**, 259 (1933); M. E. L. Oliphant, A. E. Kempton, E. Rutherford, *ibid.* **150A**, 241 (1935).
59. R. L. Cool, L. Madansky, O. Piccioni, *Phys. Rev.* **93**, 637 (1954); see also references in R. F. Peierls [*ibid.* **118**, 325 (1959)].
60. M. Alston, L. W. Alvarez, P. Eberhard, M. L. Good, W. Graziano, H. K. Ticho, S. G. Wojcicki, *Phys Rev. Lett.* **5**, 520 (1960).
61. ———, *ibid.* **6**, 300 (1961).
62. ———, *ibid.*, p. 698.
63. R. Hofstadter, *Rev. Mod. Phys.* **28**, 214 (1956).
64. W. Holladay, *Phys. Rev.* **101**, 1198 (1956); Y. Nambu, *ibid.* **106**, 1366 (1957); G. F. Chew, *Phys. Rev. Lett.* **4**, 142 (1960); W. R. Frazer and J. R. Fulco, *Phys. Rev.* **117**, 1609 (1960); F. J. Bowcock, W. N. Cottingham, D. Lurie, *Phys. Rev. Lett.* **5**, 386 (1960).
65. B. C. Maglić, L. W. Alvarez, A. H. Rosenfeld, M. L. Stevenson, *Phys. Rev. Lett.* **7**, 178 (1961).
66. J. A. Anderson, V. X. Bang, P. G. Burke, D. D. Carmony, N. Smitz, *ibid.* **6**, 365 (1961).
67. A. R. Erwin, R. March, W. D. Walker, E. West, *ibid.*, p. 628.
68. C. Perrier and E. Segrè, *Accad. Naz. Lincei, Rendi. Classe Sci. Fis. Mat. Nat.* **25**, 723 (1937).
69. A. Pevsner, R. Kraemer, M. Nussbaum, C. Richardson, P. Schlein, R. Strand, T. Toohig, M. Block, A. Engler, R. Gessaroli, C. Meltzer, *Phys. Rev. Lett.* **7**, 421 (1961).
70. P. L. Bastien, J. P. Berge, O. I. Dahl, M. Ferro-Luzzi, D. H. Miller, J. J. Murray, A. H. Rosenfeld, M. B. Watson, *ibid.* **8**, 114 (1962).
71. M. Ferro-Luzzi, R. D. Tripp, M. B. Watson, *ibid.*, p. 28.
72. G. M. Pjerrou, D. J. Prowse, P. Schlein, W. E. Slater, D. H. Stork, H. K. Ticho, *ibid.* **9**, 114 (1962).
73. L. Bertanza, V. Brisson, P. L. Connolly, E. L. Hart, I. S. Mittra, G. C. Moneti, R. R. Rau, N. P. Samios, I. O. Skillicorn, S. S. Yamamoto, M. Goldberg, L. Gray, J. Leitner, S. Lichtman, J. Westgard, *ibid.*, p. 180.
74. M. Gell-Mann, California Institute of Technology Synchrotron Laboratory Report CTSL-20, 1961 (unpublished).
75. Y. Ne'eman, *Nucl. Phys.* **26**, 222 (1961).
76. V. E. Barnes, P. L. Connolly, D. J. Crennell, B. B. Culwick, W. C. Delaney, W. B. Fowler, P. E. Hagerty, E. L. Hart, N. Horwitz, P. V. C. Hough, J. E. Jensen, J. K. Kopp, K. W. Lai, J. Leitner, J. L. Lloyd, G. W. London, T. W. Morris, Y. Oren, R. B. Palmer, A. G. Prodell, D. Radojičić, D. C. Rahm, C. R. Richardson, N. P. Samios, J. R. Stanford, R. P. Shutt, J. R. Smith, D. L. Stonehill, R. C. Strand, A. M. Thorndike, M. S. Webster, W. J. Willis, S. S. Yamamoto, *Phys. Rev. Lett.* **12**, 204 (1964).
77. A. H. Rosenfeld, A. Barbaro-Galtieri, W. J. Podolsky, L. R. Price, P. Söding, C. G. Wohl, M. Ross, W. J. Willis, *Rev. Mod. Phys.* **39**, 1 (1967).

Fourier Analysis and the Structure of DNA

Electron density functions in structure analysis are discussed, with particular reference to DNA.

Jerry Donohue

Most of the early work on the investigation of the structure of DNA was based on the calculation of the transforms of assumed molecular models and comparison of these with the observed x-ray data, together with adjustments of the models in order to improve the agreement (*1*, *2*). More recently, however, the emphasis appears to have shifted to the Fourier method to obtain proof of the structure (*3–9*). In this method, the electron density distribution is calculated by use of the following formula:

$$\rho(xyz) = \frac{1}{V}\sum_{hkl=-\infty}^{\infty} |F_{hkl}| \cos\{2\pi(hx + ky + lz) - \alpha_{hkl}\}$$

Although this formula may be used to calculate the electron density at any desired point (*xyz*) in the unit cell, its application is not straightforward. Although the structure amplitudes $|F_{hkl}|$ are observable, their phase angles α_{hkl} are unobservable; the structure amplitudes are proportional to the square root of the observed intensities, but the phase angles must be calculated from the atomic coordinates of an assumed model. If the structure contains a center of symmetry on the origin, the values which the phase angles may assume are limited to either zero or π (*10*), and the problem is thus reduced to finding the correct signs to be attached to the structure amplitudes. In the case of structures without centers of symmetry, the phases may assume any values from zero to 2π. All of the examples discussed below fall into this latter category.

The author, a crystallographer, is professor of chemistry in the University of Pennsylvania, Philadelphia.

Evidence for the Structure of DNA Based on Fourier Syntheses

Before examining the nature of the electron density maps which have been offered as proof of the existence of Watson-Crick pairing in DNA, it is first necessary to examine the nature of the data used to calculate these maps. There have been various statements regarding the extent of the experimental data which have been used in the Fourier analysis of the structure of DNA. Wilkins (*3*) stated that diffraction at angles corresponding to spacings as small as 1.1 Å had been recorded but not yet analyzed, but he also remarked "The x-ray data cannot resolve spacings less than 3 Å." He presented three electron density functions. Arnott (*6*) presented numerous Fourier diagrams; the reference for them (*4*) is an abstract of a paper presented at an international congress, which states that data out to 1.7 Å had been obtained. Arnott (*6*) also referred to earlier work (*8*) of Marvin, Wilkins, and Hamilton (actually published later, in 1966) as extending to 2.5 Å. Meanwhile, Arnott, Wilkins, Hamilton, and Langridge (*7*) published eight different electron density maps, all but one of which had been included in the earlier paper of Arnott (*6*). The paper of Marvin, Wilkins, and Hamilton (*8*) contains four Fourier plots, two of which are identical to those published 5 years earlier by Wilkins (*3*); they stated that their data included all spacings larger than 2.5 Å. The review of Hamilton (*9*) included two electron density maps previously published by Arnott (*6*), although this paper was not cited by Hamilton. He stated that the Fourier studies of Arnott *et al.* (*7*) were made with sharp reflections extending to spacings as small as 1.7 Å.

The question of the extent of the data used is an important one because of the resolution to be expected in the electron density maps. It should be pointed out that there has been confusion between resolving power and minimum observed spacing d_{min}. For example, Wilkins (*3*) and Marvin *et al.* (*8*), appear to use the two terms interchangeably. Actually, the Fourier method is, in theory, capable of resolving two peaks if they are more than 0.6 d_{min} apart (*11*). Thus, data extending to 1.7 Å should resolve atoms slightly greater than 1 Å apart. In practice the resolution may be somewhat worse than this because of errors in the observed data, or, as in the case of DNA, because of disorder. Furthermore, since the majority of the spacings of the reflections used in the Fourier syntheses (*6, 7, 9*) are greater than 3 Å, the theoretical resolution is not likely to be much better than 2 Å (*12*).

A typical electron density map which was prepared with the observed intensity data from DNA is shown in Fig. 1. The resolution is seen to be quite poor, as expected. On the basis of this and other similar maps various conclusions have been drawn, such as: "There is no indication in the syntheses . . . that the average base pair should be made larger . . . therefore it is unlikely that the Donohue scheme exists in DNA" (*3*); "examination of the Fourier syntheses also shows that the Hoogsteen scheme almost certainly cannot exist in DNA" (*3*); "the diffraction from DNA is not compatible with a Hoogsteen type of base pairing but only with one of the Watson-Crick kind" (*6*); "Fourier syntheses where the phases were provided by Hoogsteen DNA models seemed compelling proof that a model of the Watson-Crick type was the unique solution to the problem of the DNA structure" (*6*); "a base-pairing which is not of the Watson-Crick kind does not participate to any great extent in the structure of double helical DNA's" (*7*); "only a model of the Watson-Crick kind was compatible with the diffraction data" (*7*); "the [Fourier] syntheses confirm the correctness of the model" (*8*); and "they [Fourier syntheses] have excluded any significant participation of base pairs other than those of the Watson-Crick type from the DNA structure" (*9*).

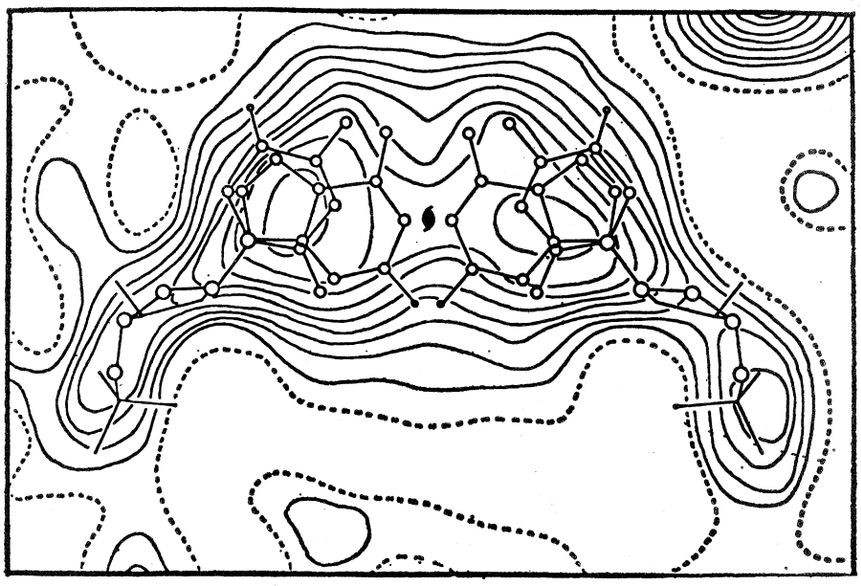


Fig. 1. Fourier synthesis calculated with the observed data from DNA, showing the distribution of electron density in the plane of a base pair. Some of the atoms of the assumed model which were used to calculate the phases are shown as open circles; the others do not lie near the plane of this section. (From reference *5*; closely similar maps may be found in refernces *3, 6–9.*)

Calculations with Synthetic Structures

To assess the validity of conclusions such as these it is necessary to test the importance of the phases, which are obtained from assumed models, to the appearance of electron density functions. To this end, three different synthetic structures, termed W, D, and H, were constructed. Each consists of a triclinic unit cell (*13*) containing a 9-methyladenine molecule hydrogen-bonded to a 1-methylthymine molecule. The structures differ in the manner of hydrogen bonding. The phases and amplitudes were calculated (*14*) for all (*hkl*) reflections having spacings greater than 2.0 Å. In order to simulate the nature of the observed DNA data and to avoid nonconvergence effects, the amplitudes were multiplied by an artificial temperature factor of exp $(-30 \sin^2\theta/\lambda^2)$ before calculation of the Fourier syntheses. These amplitudes were then treated as "observed" data in the calculation of a number of electron density functions; these data, unlike actual experimental data, are free of both random and systematic errors.

The amplitudes and phases of pairing W were used to prepare a map of the electron density projected down the *c*-

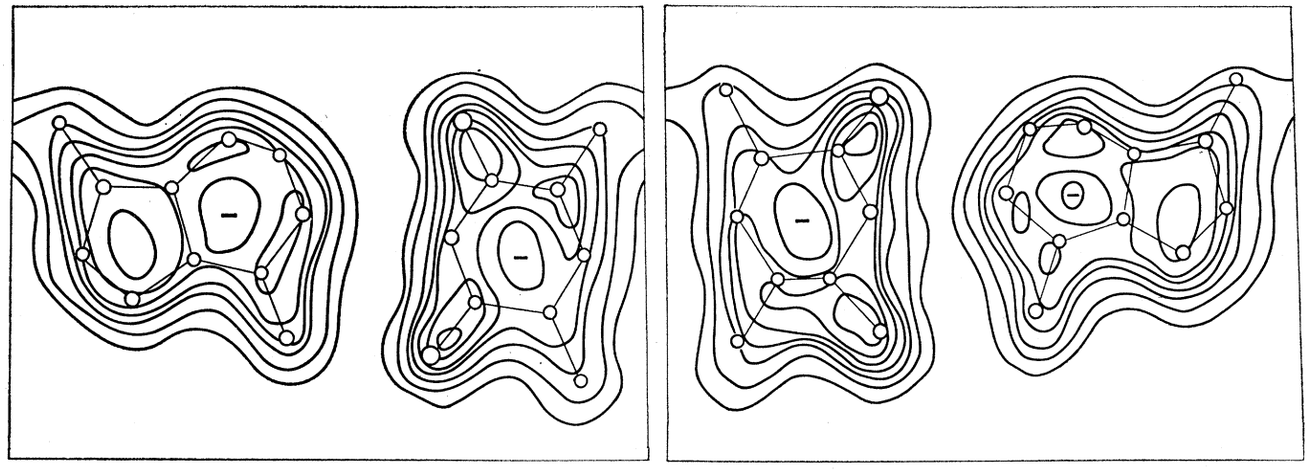


Fig. 2 (left). Electron density function of synthetic adenine-thymine pair W, calculated with the amplitudes and phases of that pair. Fig. 3 (right). Electron density function of synthetic adenine-thymine pair D, calculated with the amplitudes and phases of that pair. The positions of the adenine and thymine have been reversed with respect to Fig. 2.

axis. The result is shown in Fig. 2. This map is very similar in appearance to Fig. 1, except that the resolution is slightly better, probably because no disorder has been introduced; the simulation of DNA data by this synthetic structure is, however, verified.

The same procedure was then carried out with the amplitudes and phases of pairing D. The result is shown in Fig. 3, which bears a superficial resemblance to Fig. 2, except that the positions of the purine and pyrimidine have been reversed. Both Figs. 2 and 3, however, give good qualitative representations, at low resolution, of the structures which were used in the respective calculations. It is worth noting that the electron density falls to minima near the centers of the six-membered rings.

The importance of the phases was next tested by the preparation of an electron density map in which the amplitudes of pairing W and the phases of pairing D were used. The result is shown in Fig. 4. This is tantamount to a situation in which a structure with pairing W furnishes the observed intensities but the structure is thought to be pairing D, with the phases calculated with that model. The resulting electron density gives very little, if any, indication that the model being tested is incorrect—the Fourier series has reproduced quite faithfully the electron density of the assumed model. Figure 4 bears a close resemblance to Fig. 3, even to the appearance of the minima near the centers of the six-membered rings of the model used to obtain the phases, rather than the structure which gave the observed intensities.

Further tests were then made by making parallel calculations with the very different pairing H. Figure 5 is the electron density as calculated with both the amplitudes and phases of pairing H, and, as expected, a good qualitative representation of that structure is obtained. However, when the amplitudes of pairing W are combined with the phases of pairing H, an almost equally good representation of pairing H is obtained, as shown in Fig. 6. Again, there are no clear indications that the model being tested (H) is unsatisfactory, nor that the true structure, that is, the one corresponding to the observed intensities (W), is to be preferred.

It might be thought that this failure of the Fourier method in the above tests to reject an incorrect hypothesis resulted from the use of low-resolution data. This conjecture, in fact, is untrue, and of a number of examples included in a recent discussion (*15*) of incorrect structures for which Fourier evidence had been published, the example of tyrosine (*16*) is particularly interesting. In that work Ramachandran and Srinivasan also studied the importance

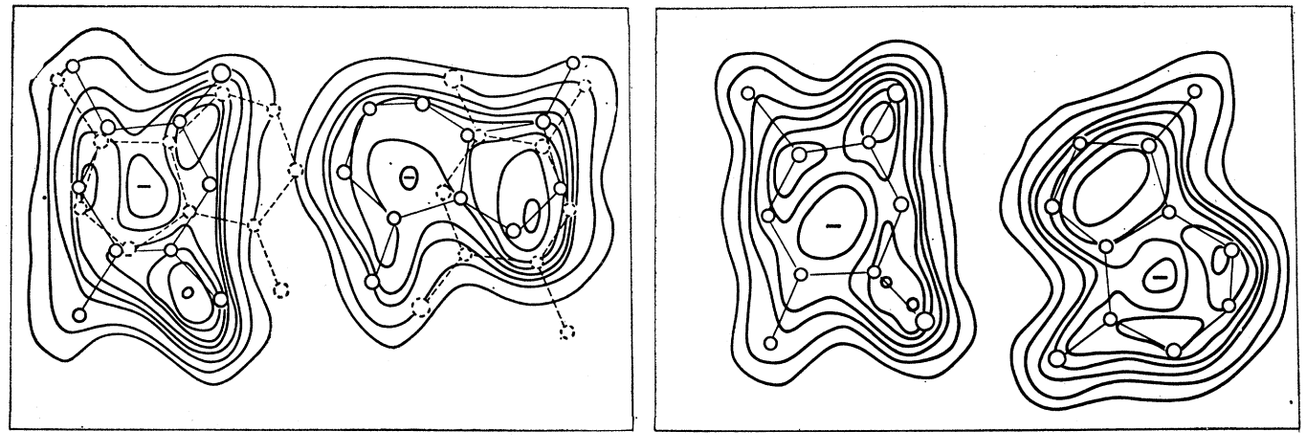


Fig. 4 (left). Electron density function calculated with the amplitudes of pairing W (dotted circles) and the phases of pairing D (full circles). Fig. 5 (right). Electron density function of synthetic adenine-thymine pair H, calculated with the amplitudes and phases of that pair.

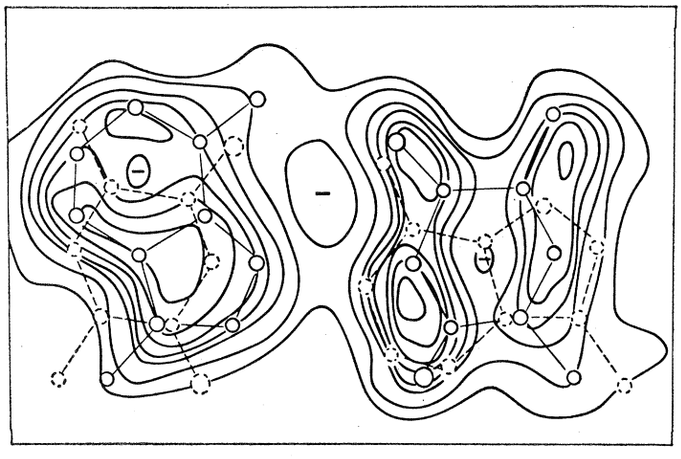
of the phases in calculations of electron density. When the amplitudes and phases of a tyrosine structure were used to calculate an electron density projection a faithful representation of the structure was, of course, obtained, as shown in Fig. 7a. They then constructed a hypothetical isoelectronic six-atom random structure and calculated the amplitudes for it. When these amplitudes were combined with the phases of the tyrosine structure, the resulting Fourier map, Fig. 7b, shows the tyrosine structure rather nicely and gives scarcely any indication of the six-atom structure which gave the "observed" amplitudes. Again, the overwhelming importance of the phases of an assumed model is strikingly demonstrated.

Further evidence for this importance is found in Fig. 8, which is an electron density function calculated with *constant* amplitudes and the phases of pairing D. It may be seen that the use of such amplitudes together with the phases of an assumed structure gives a quite recognizable image of that assumed structure. This result is quite similar to one obtained earlier by Dunitz (*17*), who calculated one of the projections of oxalic acid dihydrate using the phase angles of the correct structure and amplitudes equal to the average atom form factor at the scattering angle of each respective F. This procedure gave a rather good picture of the molecule in its correct position in the unit cell.

There are some precautionary features of Figs. 7b and 8 which suggest that all is not well, but these features do not occur in the low-resolution maps, Figs. 4 and 6. These include: (i) irregularly shaped resolved atoms, (ii) unequal peak heights for resolved equal atoms, and (iii) false detail (which cannot be explained away by solvent molecules).

Comments

It should come as no surprise that the Fourier maps calculated with the observed amplitudes from DNA and the phases of a structure with Watson-Crick pairing should recognizably reproduce that pairing, as seen in Fig. 1. On the other hand, it is clear that the conclusions cited above (*3*, *6–9*) are unjustified. While it is well known that the gross features of the Watson-Crick model are compatible with the x-ray data, the Fourier method of structure refinement has, in fact, contributed nothing toward either the proof of that structure, nor toward the elucidation of its details, because of the "pulling-yourself-up-by-your-bootstraps" aspect of that method. This property has been amply demonstrated above and has been recognized, in some quarters at least, before; for example, "Fourier synthesis, however, is not a good test



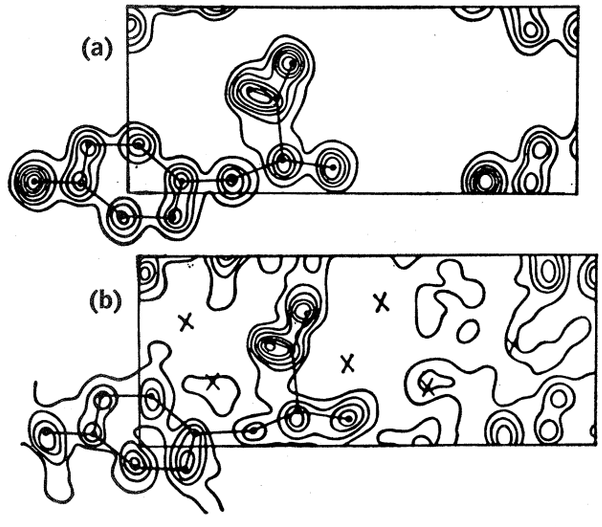


Fig. 6 (top left). Electron density function calculated with the amplitudes of pairing W (dotted circles) and the phases of pairing D (full circles).

Fig. 7 (top right). Electron density of tyrosine projected down the *c*-axis (from reference *16*, with permission of the editor of *Nature*). Phases of the correct structure, indicated by the dots, used to calculate both projections. (a) Amplitudes of the correct structure used, and (b) amplitudes of the six-atom random structure, indicated by the crosses, used.

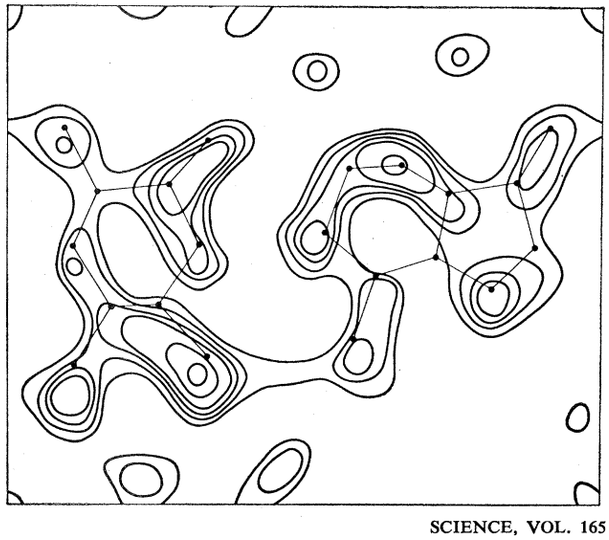


Fig. 8 (right). Electron density map calculated with equal amplitudes and the phases of pairing D.

of a proposed structure; it always tends to support the hypothesis upon which it is based" (*18*), or "however, the beauty of these maps [that is, those in (*3*)] should not overshadow the intrinsic pitfalls of noncentric structures" (*19*), or "we have observed that peaks quite comparable in height with true atomic peaks can be introduced by phasing into the electron density distribution at points where no true atoms exist" (*20*).

Conclusions

It is obvious that the published x-ray diffraction data from DNA are incapable of being analyzed by the Fourier method for the purpose of obtaining evidence as to what the structure of that substance is, not to mention refining a proposed model. It is, accordingly, not inappropriate to ask just what the nature of such data should be in order that calculations of electron density could yield conclusive results. In my opinion, there are a number of problems which will have to be surmounted before this end can be achieved.

1) *Concerning the resolution problem*, it seems probable that the data used up to now in the calculation of electron density functions for DNA extend to spacings of only about 3 Å. Clearly, this is not far enough. The data should be capable of resolving peaks corresponding to pairs of bonded atoms. This means that data must be collected almost to the limit of CuKα radiation *in all directions*, not just to the higher orders of the very strong 3.4 Å reflection which presumably arises from the stacking of the bases. Thus, specimens of DNA which exhibit a higher degree of crystallinity than have been examined will have to be prepared.

2) *The disorder problem.* The disorder and resolution problems are seriously interconnected. It will not ever be possible to obtain data from DNA which are capable of resolving peaks corresponding to certain of the atoms in a disordered model (Fig. 1). Consequently, even if highly crystalline preparations do become available, there remains the intrinsic disorder of the molecule itself which arises from the lack of short-range periodicity of the bases along the polynucleotide chain. Electron density functions of these portions of the molecule will always be ill-defined smears, and the particular structure of these will have to be arrived at from secondary considerations, such as unequivocal positioning of the atoms in the sugar-phosphate backbone plus the nitrogen atoms in the glycosidic bonds.

3) *The phase problem.* Clearly, calculation of electron density functions with phases derived from an assumed structure cannot furnish proof of that structure. Evaluation of the phases must therefore be sought by other means. Because of the complexity of the DNA structure, it appears that the so-called direct methods (*21*) for the determination of phase angles will probably be ineffective (*22*). Moreover, even if the complex nature of the DNA structure is substantially simplified by the presence of noncrystallographic symmetry elements, the intensity statistics on which the direct methods depend are not valid for low-resolution data (*23*).

Phase angles for acentric structures can be deduced by careful analysis of diffraction data from a parent compound and at least two strictly isomorphous heavy atom derivatives (*24*). This method has had notable success, for example, in the case of the proteins hemoglobin and myoglobin (*25*). Whether or not similarly useful derivatives of DNA can be prepared is another question.

4) *The agreement problem.* The final test for the correctness of any structure lies in the agreement between the observed and calculated structure amplitudes. It is conventional for crystallographers to quote R values (*26*) for their refined structures. Unfortunately, apparently acceptable R values of around 20 percent have been obtained for grossly incorrect structures (*27*), and values as low as 7 percent have been reported for structures with incorrect details, that is, "partially correct" structures (*28*). Thus, achievement of an acceptable overall R factor is insufficient: the *individual* observed and calculated amplitudes must be examined, with particular attention being paid to any serious and possibly systematic discrepancies. The overall R value reported for the A form of sodium DNA is 39 percent (*2*); comparison of the individual values of the amplitudes is not possible because the customary table of the observed and calculated values for these has not been published.

Summary

Examination of the Fourier method of crystal structure analysis, in which the distribution of electron density is calculated with the observed structure amplitudes combined with phase angles obtained from an assumed model, leads to the not unexpected conclusion that proof of structure cannot be obtained in this way, particularly when only low-resolution data are available.

References and Notes

1. M. H. F. Wilkins, A. R. Stokes, H. R. Wilson, *Nature* **171**, 738 (1953); R. E. Franklin and R. G. Gosling, *ibid.*, p. 740; M. H. F. Wilkins, W. E. Seeds, A. R. Stokes, H. R. Wilson, *ibid.* **172**, 759 (1953); M. Feughelman, R. Langridge, W. E. Seeds, A. R. Stokes, H. R. Wilson, C. W. Hooper, M. H. F. Wilkins, R. K. Barclay, L. D. Hamilton, *ibid.* **175**, 834 (195➡ R. Langridge, W. E. Seeds, H. R. Wilson, C. W. Hooper, M. H. F. Wilkins, L. D. Hamilton, *J. Biophys. Biochem. Cytol.* **3**, 767 (1957); R. Langridge, D. A. Marvin, W. E. Seeds, H. R. Wilson, C. W. Hooper, M. H. F. Wilkins, L. D. Hamilton, *J. Mol. Biol.* **2**, 38 (1960); D. Marvin, M. Spencer, M. H. F. Wilkins, L. D. Hamilton, *ibid.* **3**, 547 (1961).
2. W. Fuller, M. H. F. Wilkins, H. R. Wilson, L. D. Hamilton, *J. Mol. Biol.* **12**, 60 (1965).
3. M. H. F. Wilkins, in *Biological Structure and Function*, T. W. Goodwin and O. Lindberg, Eds. (Academic Press, New York, 1961), p. 13.
4. S. Arnott, M. H. F. Wilkins, D. A. Marvin, R. Langridge, A. Rich, L. D. Hamilton, *Acta Cryst.* **16**, A77 (1963).
5. M. H. F. Wilkins, *Science* **140**, 941 (1963).
6. S. Arnott, *Bull. Inst. Phys. Soc.* **15**, 169 (1964).
7. S. Arnott, M. H. F. Wilkins, L. D. Hamilton, R. Langridge, *J. Mol. Biol.* **11**, 391 (1965).
8. D. A. Marvin, M. H. F. Wilkins, L. D. Hamilton, *Acta Cryst.* **20**, 663 (1966).
9. L. D. Hamilton, *Nature* **218**, 633 (1968).
10. For a lucid explanation of how this limitation comes about, see J. Waser, *J. Chem. Educ.* **45**, 446 (1968).
11. W. L. Bragg and J. West, *Phil. Mag.* **10**, 823 (1930); C. W. Bunn *Chemical Crystallography* (Oxford Univ. Press, London, 1945), p. 350.
12. S. Arnott, private communication.
13. These unit cells all have dimensions $a = 12.50$, $b = 10.41$, $c = 3.40$Å, and $\alpha = \beta = \gamma = 90°$.
14. In these calculations the atom form factor curves for carbon, nitrogen, and oxygen were taken from *International Tables for X-Ray Crystallography*; the scattering by the hydrogen atoms was neglected.
15. J. Donohue, material presented at the symposium, on *Aims of Increased Accuracy in Diffraction Problems* (State Univ. of New York at Buffalo, March 1969).
16. G. N. Ramachandran and R. Srinivasan, *Nature* **190**, 159 (1961).
17. J. D. Dunitz, private communication, 1949.
18. P. R. Pinnock, C. A. Taylor, H. Lipson, *Acta Cryst.* **9**, 173 (1956).
19. V Luzzati, *Progr. Nucleic Acid Res. Mol. Biol.* **1**, 347 (1963).
➡ D. C. Hodgkin, J. Pickworth, J. H. Robertson, R. J. Prosen, R. A. Sparks, K. N. Trueblood, *Proc. Roy. Soc. Ser. A* **251**, 306 (1959).
21. M. M. Woolfson, *Direct Methods in Crystallography* (Clarendon Press, Oxford, 1961).
22. M. F. Perutz and V. Scatturin, *Acta Cryst.* **7**, 799 (1954).
23. R. B. Corey, J. Donohue, K. N. Trueblood, K. J. Palmer, *ibid.* **8**, 518 (1955); D. Harker, *ibid.* **6**, 731 (1953).
24. D. Harker, *ibid.* **9**, 1 (1956); M. F. Perutz, *ibid.*, p. 867.
25. M. F. Perutz, Science **140**, 863 (196➡ J. C. Kendrew, *ibid.* **139**, 1259 (1963).
26. The function R is the average percentage discrepancy between the observed structure

amplitudes and those calculated on the basis of a proposed structure: $R = 100\ (\Sigma||F_{obs}| - |F_{calc}||)/\Sigma|F_{obs}|$. For structures refined on the basis of visually estimated intensity data, R generally ranges between about 8 to 30 percent, depending on how thorough the refinement is and, of course, on the accuracy of the data. In more recent structure refinements based on diffractometer data, the values of R achieved are usually less than 10 percent and may be as low as 2 percent.

27. J. Donohue and K. N. Trueblood, *Acta Cryst.* **9**, 615 (1956).
28. J. Donohue and S. H. Goodman, *ibid.* **22**, 352 (1967); R. Parthasarathy, *Science* **161**, 179 (1968).

29. Supported by the Advanced Research Projects Agency, Office of the Secretary of Defense. I benefited from discussion, criticism, and advice from Drs. Max Dobler, J. D. Dunitz, Neville Kallenbach, Richard E. Marsh, Linus Pauling, Bryan W. Roberts, Stephen L. Schwartz, Kenneth N. Trueblood, and Jürg Waser. Mrs. Maryellin Reinecke prepared many of the figures; Dr. H. L. Carrell and Mr. F. Soule carried out some of the calculations.

Academic Research in Germany: A New Support Program

National interest, university welfare, and research needs are combined in one program.

Bernard R. Stein

An important shift in the thinking governing the support of academic research in the Federal Republic of Germany was revealed in a report issued in July 1967 by the *Wissenschaftsrat* (Science Council), a major governmental advisory group (*1*). The report, dealing with the planned expansion of the universities until 1970, recommended establishment of a *Sonderforschungsbereiche* (special research areas) program to support the formation of cooperative, multidisciplinary research efforts. Such efforts, although located within universities, would also provide for the involvement of individuals and groups outside the academic community. Three criteria would determine the selection of any special research area: intellectual or scientific merit, university considerations, and the national interest. The group or groups identified with each criterion would participate in the decisions.

In July 1967 the *Deutsche Forschungsgemeinschaft* (German Research Association), the principal national organization responsible for the support of academic research, agreed to advise the Science Council on the selection of special research areas proposals and manage the operations of the program. Details on proposal submission and selection procedures, implementation and financial arrangements, and the first list of approved research areas were revealed in a document issued by the Science Council in July 1968 (*2*). The list contains 141 areas divided among seven categories as follows: disciplines of the philosophical faculties, 8; regional area studies, 11; law, economics, and social sciences, 10; general and dental medicine, 35; mathematics and natural sciences, 37; veterinary medicine, 4; and engineering and architecture, 36. Among the 18 areas already funded at a total cost of slightly more than $1 million are the following: Southeast Asia research (Heidelberg), synoptic meteorology (Berlin), molecular basis of development (Freiburg), and medical statistics and documentation (Mainz) (*3*).

Proposal Submission and Selection Procedures

Under the research areas program a group of university researchers may submit a collective research proposal to the principal university decision-making body. In most cases this will probably be the senate or one of its authorized committees. Those proposals which are approved are then forwarded for comment to the appropriate state ministry of education (*Kultusministerium*), the agency which bears primary responsibility for the welfare of the university. In the Federal Republic there is no federal ministry of education, the education function having been assigned to the eleven Länder or states in the postwar settlement (*4*). The proposals are then submitted to the Science Council which routes them to the *Deutsche Forschungsgemeinschaft*, the German equivalent of the U.S. National Science Foundation, for review and evaluation according to guidelines established by the former. This evaluation, prepared with the help of the most competent scholars, carries great weight in the final decisions made by the Science Council. The Council may reject a favorable recommendation by the German Research Association, but may not, on its own, list any proposal for a special research area not previously approved by the Research Association or the appropriate state ministry of education.

Although the principal initiative for proposing a special area lies with the universities, the federal government, individual states, the Max Planck Society, and the Research Association may also recommend candidate proposals. Each must be submitted to the Science Council accompanied by a statement identifying goals and, if possible, specifying the university and the particular groups considered most appropriate to undertake the research. Following discussion by the Science Council, the proposal will be examined by the appropriate Association committee, and, if approved, the suggested university and respective state ministry of education are invited to present their views. If all parties agree that the proposal is in their interest, a formal application may be submitted and processed in accordance with the above procedure.

Only those special research areas appearing on the Science Council list may be considered for support from special federal and state funds. On the basis of research area priorities established by the Science Council with the aid of the Research Association, the universities are invited to submit requests for funds. Funding decisions are then made by a Research Association committee specifically constituted for this purpose.

The author is a staff associate in the Office of Planning and Policy Studies of the National Science Foundation, Washington, D.C. 20550.